Abstract No. Guo0308

Crystal Structure of the Heterodimeric Complex of the Adaptor, ClpS, with the N-domain of the AAA$^+$ Chaperone, ClpA

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Beamline(s): X9B

Introduction: ClpA, an Hsp100/Clp chaperone and regulator of ClpAP protease complex, participates in post-translational protein quality control and regulation and modulated by an adaptor protein, ClpS, through its interaction with the N-domain of ClpA.

Methods and Materials: The N-domain of ClpA and ClpS were overexpressed and purified to homogeneity, respectively. The complex of the N-domain and ClpS (ClpNS) was obtained by one step gel filtration chromatography of the equal molar ratio mix of the N-domain and ClpS and further crystallized in two forms. The x-ray diffraction data were collected and the structures were solved.

Results: The crystal structures of protein complex ClpNS were solved in two different forms at 2.3 and 3.3 Å, respectively. The ClpS structure forms a α/β-sandwich and is topologically analogous to the C-terminal domain of the ribosomal protein L7/L12. ClpS contacts two surfaces on the N-terminal domain in both crystal forms; the more extensive interface was shown to be favored in solution by protease protection experiments. The N-terminal 20 amino acid residues of ClpS are invisible in crystal; the ClpSΔ17, in which the first 17 residues were removed, binds the ClpA N-domain and no longer inhibits ClpA activity. A zinc-binding site involving two Histidine and one Glutamate residues was identified in the N-terminal domain of ClpA. In a model of ClpS bound to hexameric ClpA, ClpS is oriented with its N terminus directed toward the distal surface of ClpA, suggesting that the N-terminal region of ClpS may affect productive substrate interactions at the apical surface or substrate entry into the ClpA translocation channel.

Acknowledgments: We thank Dr. Z. Dauter for assistance in the data collection at X9B of NSLS.

References:

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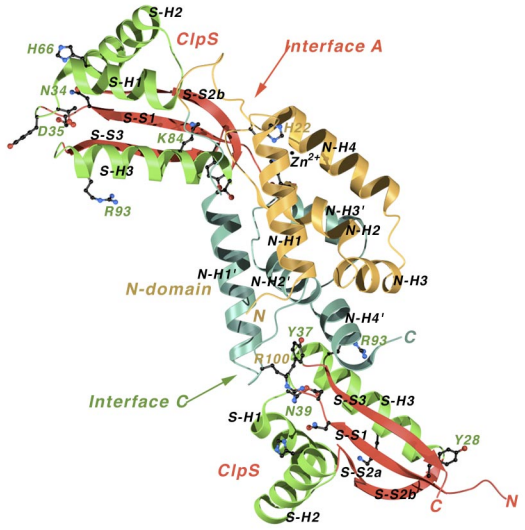

The structure of the N-domain of ClpA complexed with ClpS